OLUYEMI, G.F., AFOLABI, R.O., ZAMORA, S.C., LI, Y. and MCELROY, D. 2024. Environmental impact assessment of a plant cell-based bio-manufacturing process for producing plant natural product ingredients. *Sustainability* [online], 16(19), article number 8515. Available from: https://doi.org/10.3390/su16198515

Environmental impact assessment of a plant cell-based bio-manufacturing process for producing plant natural product ingredients.

OLUYEMI, G.F., AFOLABI, R.O., ZAMORA, S.C., LI, Y. and MCELROY, D.

2024

sustainability

MDPI

Article

Environmental Impact Assessment of a Plant Cell-Based Bio-Manufacturing Process for Producing Plant Natural Product Ingredients

Gbenga F. Oluyemi [1,*], Richard O. Afolabi [1], Samuel Casasola Zamora [2], Yuan Li [2,*] and David McElroy [2]

[1] School of Engineering, Robert Gordon University, Aberdeen AB10 7GJ, UK; r.afolabi@rgu.ac.uk
[2] Green Bioactives Limited (GBL), Douglas House, Pentlands Science Park, Penicuik, Edinburgh EH26 0PL, UK; samuel.casasola@greenbioactives.com (S.C.Z.); david.mcelroy@greenbioactives.com (D.M.)
* Correspondence: g.f.oluyemi@rgu.ac.uk (G.F.O.); yuan.li@greenbioactives.com (Y.L.)

Abstract: Purpose: This study employed a Life Cycle Assessment (LCA) methodology to evaluate the environmental impacts of a novel plant cell-based biomanufacturing process for producing plant natural product ingredients. The primary purpose was to assess the relative sustainability of the process and to provide insights into potential areas of improvement in the biomanufacturing process. Method: The LCA method used an MS Excel (Ver. 2407) -based approach with a cradle-to-gate system boundary covering raw material sourcing (A1), raw material transportation (A2), and product extract manufacturing (A3) stages. Energy use and material inventory data are presented for different unit operations, and environmental impact factors were obtained from the Ecoinvent database. The study included a Material Circularity Index (MCI) calculation to assess the circularity of the biomanufacturing process for the production of saponin emulsifiers that are normally extracted from the woody tissue of the Chilean soapbark tree (*Quillaja saponaria*). Comparative analyses were performed against a wild-harvest approach for plant tannin extraction from spruce (*Picea abies*) tree bark. Key Results: The environmental impact assessment focused on determining relative Global Warming Potential (GWP), Acidification Potential (AP), Freshwater Eutrophication (FE), Particulate Matter Formation (PMF), and Ozone Depletion Potential (ODP). Results indicated that the extract manufacturing stage (A3) contributed significantly to adverse environmental impacts, with varying levels of effects based on the energy source used. Comparative analysis with the wild harvest approach highlights the lower environmental impact of the alternative biomanufacturing process. The biomanufacturing process showed a 23% reduction in GWP, AP, and FE and a 25% reduction in PMF and ODP relative to the wild harvest approach. However, the MCI for the biomanufacturing process was estimated to be 0.186, indicating a low material circularity. Conclusions: The results revealed that the extract manufacturing stage, particularly energy consumption, significantly influences the relative environmental impacts of the alternative production processes. Different energy sources exhibit varying effects, with renewable energy sources showing lower environmental impacts. The Material Circularity Index indicated a low circularity for the biomanufacturing process, suggesting opportunities for improvement, such as incorporating recycled or reused materials. Compared with the tannin extraction process, the plant cell-based biomanufacturing process demonstrated lower environmental impacts, emphasising the importance of sustainable practices and the use of renewable energy sources in future plant natural product sourcing. Recommendations include implementing more sustainable practices, optimising raw material choices, and extending product life spans to enhance circularity and overall environmental benefits.

Keywords: biomanufacturing; circularity; life cycle assessment; plant cell

check for updates

Citation: Oluyemi, G.F.; Afolabi, R.O.; Zamora, S.C.; Li, Y.; McElroy, D. Environmental Impact Assessment of a Plant Cell-Based Bio-Manufacturing Process for Producing Plant Natural Product Ingredients. *Sustainability* **2024**, *16*, 8515. https://doi.org/10.3390/su16198515

Academic Editor: Harvey Hou

Received: 21 August 2024
Revised: 23 September 2024
Accepted: 27 September 2024
Published: 30 September 2024

1. Introduction

The global demand for plant-derived natural products in diverse industries such as pharmaceuticals, cosmetics, and nutraceuticals has placed considerable pressure on traditional production methods, particularly wild harvesting. This practice, which involves the extraction of plant materials directly from natural ecosystems, has long served as a primary source for phytochemicals and other plant-derived bioactive compounds. However, the growing scale of demand for plant-derived natural products, coupled with the environmental and social costs of wild harvesting and climate change impacts, has raised significant sustainability and availability concerns around future product accessibility [1,2]. Traditional wild harvesting can lead to biodiversity loss, habitat degradation, and the depletion of natural resources, ultimately threatening the stability of ecosystems and the livelihoods of communities dependent on them [3–6]. These vulnerabilities are further exacerbated by the impacts of climate change and the resulting increased unpredictability of affordable raw material availability [7].

In response to these challenges, plant cell culture (PCC) technology has emerged as a promising alternative for the production of phytochemicals [8–10]. Unlike traditional wild harvesting, PCC enables the controlled in vitro cultivation of plant cells, allowing for the year-round production of high-value plant-derived secondary metabolites without the need for extensive land use or resource extraction from natural habitats [11–13]. This alternative biomanufacturing approach offers several advantages, including the ability to produce specific phytochemicals year-round, independent of environmental conditions such a drought or pestilence, while reducing the pressure on natural ecosystems. PCC also facilitates the production of consistent, high-quality bioactive compounds that are crucial for industries reliant on stable supply chains [13–15].

However, despite the potential environmental benefits of PCC, the energy-intensive nature of PCC-based production systems—particularly as these are non-photosynthetic heterotrophic processes—raises important questions about their overall sustainability [16,17]. To date, examples of comprehensive environmental impact assessments of PCC systems for phytochemical production remain limited [18]. This study seeks to address this gap by employing a Life Cycle Assessment (LCA) to evaluate the environmental impacts of Green Bioactives Limited's (GBL) novel plant cell culture-based biomanufacturing process for phytochemical production. LCA provides a holistic framework for assessing the environmental performance of biomanufacturing processes by analysing inputs and outputs across the entire production cycle, from raw material sourcing to the final product [19].

This study focuses on key environmental indicators, such as Global Warming Potential (GWP), Acidification Potential (AP), and Material Circularity Index (MCI), to assess the environmental performance of the plant cell culture-based biomanufacturing process. Additionally, the research compares the environmental impacts of this innovative plant cell-based approach with those of traditional wild harvesting methods, specifically for the production of tannins [20]. By examining these factors, this study aims to identify areas for improvement in the sustainability of the PCC processes, such as optimizing energy use and incorporating more circular practices. The findings will contribute to a better understanding of the environmental implications of plant cell culture technology and support the transition toward more sustainable and reliable phytochemical production methods.

2. Methodology

2.1. Life Cycle Assessment

2.1.1. Aim and Scope

The aims of this assessment were to: (1) assess and compare the environmental impact of the plant cell culture-based biomanufacturing process; (2) identify critical process stage(s) contributing to environmental load; (3) assess the circularity of the biomanufacturing process with a Material Circularity Index (MCI) calculation; and (4) compare the impact of the plant cell culture-based biomanufacturing process with a traditional wild harvest approach across different stages of manufacturing and to identify the difference in environmental impact between the two manufacturing approaches.

2.1.2. Standards, Software and Database

This LCA was conducted in accordance with ISO14040 and ISO14044 [21,22]. The various environmental impact factors were obtained from the Ecoinvent database (https://ecoinvent.org/the-ecoinvent-database/, accessed on 3 November 2023) [23] using the OpenLCA software 2.0 (https://www.openlca.org/, accessed on 3 November 2023) [24].

2.1.3. System Boundaries

The system boundary of the plant cell culture-based production process is given in Figure 1. This indicate the limits of the system being investigated in this study using the LCA method (Section 2.1.5). The following energy-consuming production stages were considered within the system boundaries, including production process, extraction, and post-extraction: the production process including upstream processing comprising inoculum and media preparation; the growth of plant cells in the bioreactor (biomanufacturing); plant cell harvesting after the biomanufacturing stage; a preprocessing stage including separation of conditioned media and biomass; and an optional drying stage to obtain dried biomass and cell disruption through homogenisation. The extraction stage included 4 scenarios to yield different concentrations of plant cell extract depending on biomass input and solvent/biomass ratio; scenario 1, 2, and 3 yielded 7.8% (*w*/*v*), 25% (*w*/*v*) and 50% (*w*/*v*) fresh cell weight (FCW) extract, respectively; and scenario 4 yielded 20% (*w*/*v*) dry cell weight (DCW) extract. The post-extraction stage included 4 approaches to yield different extracts: centrifugation of the resulting extract from extraction scenario 1, 2, or 3 yielded liquid extract 1 comprising 7.8% (*w*/*v*), 25% (*w*/*v*), or 50% (*w*/*v*) FCW. An additional spray dry process following the aforementioned centrifugation step yield dried extract in powder form. Centrifugation of the resulting extract from extraction scenario 4 yielded liquid extract 2 comprising 20% (*w*/*v*) DCW, and a 60% concentrated extract can be achieved via evaporation of liquid. The dried extract, liquid extract 2, or evaporated extract can be further diluted with solvent.

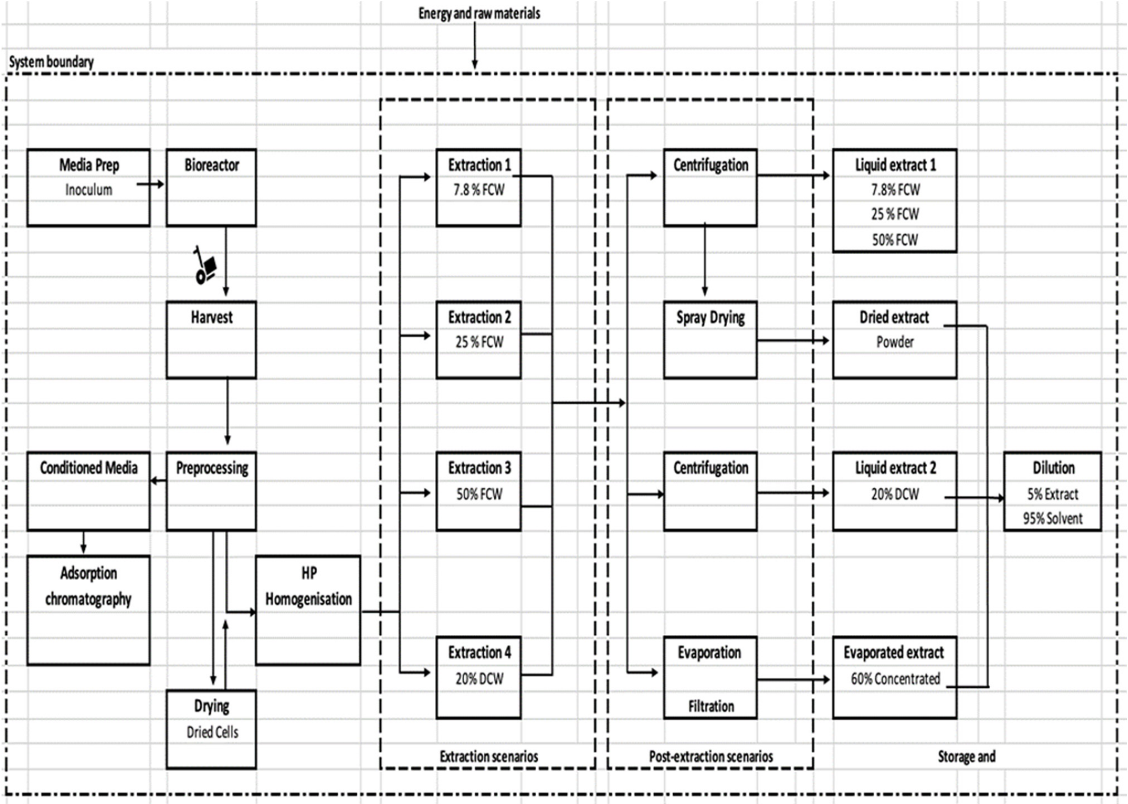

Figure 1. System boundary for the plant cell culture-based biomanufacturing process, comprising different extraction scenarios and final product formats.

2.1.4. Life Cycle Inventory (LCI)

Tables 1–4 summarise the life cycle inventory data for the material and energy requirements of the plant cell culture-based biomanufacturing process. Table 1 captures the various operations and energy requirements provided by GBL that constitute the three stages of the biomanufacturing process (production, extraction, and post-extraction concentration of the final product) to manufacture 1 kg of plant extract (with final product specification depend on the scenario). These data cover the different extraction and post-extraction scenarios highlighted in Figure 1.

Table 1. Inventory data for the energy input to the various unit operations of the biomanufacturing process.

	Biomanufacturing Processes	Description	Energy (kWh)
Production Process	Upstream Processing	Preparation of VSC inoculum and media	68.0
	Biomanufacturing	Growth of plant cells in the bioreactor	2184.0
	Homogenisation	Cell disruption in solvent	15.0
	Drying	Removal of water from the system	160.0
	Adsorption	Use of resins to recover biomolecules from conditioned media	0.0
Extraction	Extraction 1 (7.8% FCW)	Extraction	20.0
	Extraction 2 (15% FCW)	Extraction	20.0
	Extraction 3 (20% DCW)	Extraction	80.0
	Extraction 4 (50% FCW)	Extraction	20.0
Post-Extraction	Post-Extraction Scenario 1	Centrifugation Spray Drying	22.1 60.0
	Post- Extraction Scenario 2	Centrifugation	22.1
	Post-Extraction Scenario 3	Evaporation Filtration	37.5 3.6

Table 2. Embodied environmental factors of various energy sources considered in the analysis.

Energy Source	Embodied Carbon Factor (kgCO2e/kWh)	Embodied Acidification Factor (kgSO2e/kWh)	Embodied Eutrophication Factor (kgPe/kWh)	Embodied Particulate Matter Factor (kgPM2.5e/kWh)	Embodied Ozone Depletion Factor (kgCFC11e/kWh)
Energy Mix	4.66×10^{-2}	1.90×10^{-4}	1.71×10^{-5}	8.14×10^{-6}	4.20×10^{-7}
Geothermal	7.54×10^{-2}	2.60×10^{-4}	3.87×10^{-5}	1.60×10^{-4}	2.50×10^{-8}
Hydropower	4.31×10^{-3}	1.33×10^{-5}	1.50×10^{-6}	9.90×10^{-6}	4.99×10^{-7}
Wind	1.34×10^{-2}	5.88×10^{-5}	1.05×10^{-5}	3.23×10^{-5}	5.83×10^{-9}
Solar	7.84×10^{-2}	3.50×10^{-4}	5.17×10^{-5}	1.80×10^{-4}	3.35×10^{-7}

Table 2 captured the embodied environmental factors of energy resources including mixed energy comprising 70% fossil fuel sources (natural gas, coal, and oil) and 30% renewable sources (geothermal, hydropower, wind and solar), geothermal, hydropower, wind, and solar energy.

Table 3 summarises the materials, quantities, supplier/manufacturer (assumed to be in Glasgow, UK), mode of transport of materials to GBL (in this case, a lorry), and the total distance from Glasgow to GBL (Edinburgh, UK).

Table 3. Inventory data for the material input into the biomanufacturing process.

Materials	Mass Per Volume (kg/L)	Supplier Location (A)	GBL Location (B)	Mode of Transport	Distance from A to B (km)
MS Salts	0.0043	Glasgow, UK PA4 9RF	Edinburgh UK EH26 0PL	Lorry	96.1
B5 Vitamin Stock 1000X	0.0010	Glasgow, UK PA4 9RF	Edinburgh UK EH26 0PL	Lorry	96.1
Sucrose	0.0300	Glasgow, UK PA4 9RF	Edinburgh UK EH26 0PL	Lorry	96.1
Casein hydrolysate	0.0005	Glasgow, UK PA4 9RF	Edinburgh UK EH26 0PL	Lorry	96.1
PVP-10	0.0015	Glasgow, UK PA4 9RF	Edinburgh UK EH26 0PL	Lorry	96.1
6-BA	0.000001	Glasgow, UK PA4 9RF	Edinburgh UK EH26 0PL	Lorry	96.1
Kinetin	0.000001	Glasgow, UK PA4 9RF	Edinburgh UK EH26 0PL	Lorry	96.1
NAA	0.000001	Glasgow, UK PA4 9RF	Edinburgh UK EH26 0PL	Lorry	96.1
2,4-D	0.000003	Glasgow, UK PA4 9RF	Edinburgh UK EH26 0PL	Lorry	96.1
Water	1.000000	Edinburgh UK EH26 0PL	Edinburgh UK EH26 0PL	N/A	0.0

Table 4. Embodied environmental factors of materials input into the biomanufacturing process.

Material	Embodied Carbon Factor ($kgCO_2e/kWh$)	Embodied Acidification Factor ($kgSO_2e/kWh$)	Embodied Eutrophication Factor (kgPe/kWh)	Embodied Particulate Matter Factor (kgPM2.5e/kWh)	Embodied Ozone Depletion Factor (kgCFC11e/kWh)
MS salt	3.90×10^{-2}	1.40×10^{-5}	5.91×10^{-8}	0.00×10^{0}	0.00×10^{0}
B5 Vitamin stock, 1000X	1.95×10^{0}	5.87×10^{-3}	4.40×10^{-4}	2.58×10^{-3}	4.01×10^{-7}
Sucrose	0.00×10^{0}	0.00×10^{0}	0.00×10^{0}	0.00×10^{0}	0.00×10^{0}
Casein hydrolysate	2.05×10^{0}	1.07×10^{-2}	8.00×10^{-4}	4.20×10^{-3}	1.56×10^{-5}
PVP-10	1.95×10^{0}	5.87×10^{-3}	4.40×10^{-4}	2.58×10^{-3}	4.01×10^{-7}
6-BA	1.95×10^{0}	5.87×10^{-3}	4.40×10^{-4}	2.58×10^{-3}	4.01×10^{-7}
Kinetin	1.95×10^{0}	5.87×10^{-3}	4.40×10^{-4}	2.58×10^{-3}	4.01×10^{-7}
NAA	1.42×10^{0}	5.87×10^{-3}	4.40×10^{-4}	2.58×10^{-3}	4.01×10^{-7}
2,4-D	1.95×10^{0}	5.87×10^{-3}	4.40×10^{-4}	2.58×10^{-3}	4.01×10^{-7}
Water	0.00×10^{0}	0.00×10^{0}	0.00×10^{0}	0.00×10^{0}	0.00×10^{0}

2.1.5. Life Cycle Impact Assessment (LCIA) Method

An MS Excel-based LCIA approach was conducted to evaluate the relative environmental impacts of the plant cell culture-based biomanufacturing process in producing 1 kg of plant extract at four different % of fresh cell weight (FCW) (Table 1).

The LCIA was conducted using a cradle-to-gate approach, with the life cycle stages comprising of raw material sourcing (A1), transportation (A2), and production (A3). In this case, A3 deals with the energy input for the plant cell culture-based biomanufacturing process, and A1 deals with the production of the raw materials that serve as the ingredients required as inputs into that biomanufacturing process. A2 covers the transportation of the raw materials from their manufacturer or supplier to the plant cell culture-based biomanufacturing process. The following impact categories were chosen: Global Warming Potential (GWP), Acidification Potential (AP), Freshwater Eutrophication (FE), Particulate Matter Formation (PMF), and Ozone Depletion Potential (ODP). These were selected by considering previous LCA studies on the wild harvesting process for phytochemical production (tannin) [20] and for their relevance to electricity use [25].

2.2. Material Circularity Index

The material circularity indicator (MCI) [26,27] for the biomanufacturing process was determined using the following formula in (1), as per the Ellen MacArthur Foundation:

$$MCI = 1 - LFI * F(X) \tag{1}$$

LFI refers to the linear flow index, which measures the proportion of material flowing linearly, sourced from virgin materials and ending up as unrecoverable waste. The *LFI* was calculated as shown in (2):

$$LFI = \frac{V+W}{2M} \tag{2}$$

where V is the mass of virgin feedstock, W is the mass of the irrecoverable waste, and M is the mass of the product. The values for V and W were calculated as shown in (3) and (4), respectively.

$$V = M(1 - F_R - F_U - F_S) \tag{3}$$

$$W = M(1 - C_R - C_U - C_C) \tag{4}$$

where F_R, F_U, and F_S represent the fraction of feedstock derived from recycled, reused, and biological materials, respectively. In addition, C_R, C_U, and C_C represent a fraction of the product collected for recycling, reuse, and composting. The function $F(X)$ is defined as shown in (5).

$$F(X) = \frac{0.9}{X} \tag{5}$$

where X is the product utility defined as shown in (6).

$$X = \frac{L}{L_{av}} \text{ or } X = \frac{U}{U_{av}} \tag{6}$$

where L is the life span of the product, and L_{av} is the industry average life span of a comparable product. U is the number of functional units achieved during the use of the product, and U_{av} is the average number of functional units achieved for a comparable product.

3. Results and Discussion

The LCIA was conducted with a focus on five impact categories: Global Warming Potential (GWP), Acidification Potential (AP), Freshwater Eutrophication (FE), Particulate Matter Formation (PMF), and Ozone Depletion Potential (ODP). This assessment's functional unit (FU) was The Yield of Final Plant Extract (in kg at the % FCWs of extract as indicated in Table 1). The FU was selected to allow for comparison with the chosen closest wild harvest approach with available LCA data–tannin extraction from spruce (Picea abies) bark [20]. To provide a more complete proxy for comparative analysis, it was assumed that the tannin extract was produced from spruce trees grown in Chile and that the final extract was then shipped to the plant cell culture-based biomanufacturing location in Edinburgh. The system boundary for the wild harvest approach is given in Figure 2 below, and the associated inventory data showed in Table 5 [20].

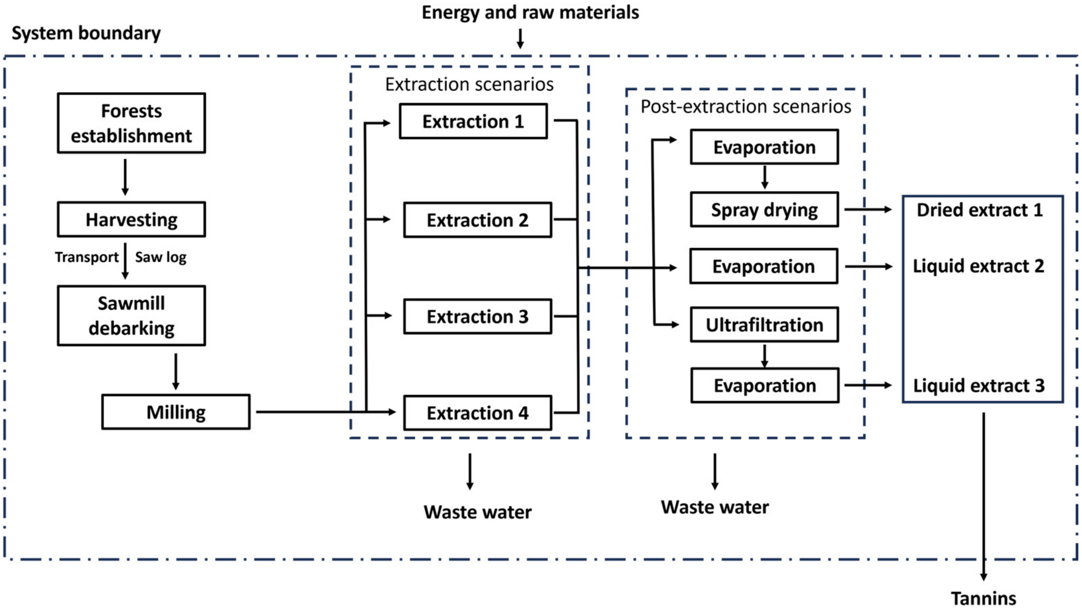

Figure 2. System boundary for the tannin extraction from spruce bark, Figure adapted from [20] licenced under CC-NC 4.0 (https://creativecommons.org/licenses/by-nc/4.0/). Modification were made. Extraction 1: One step of hot-water extraction. Extraction 2: One step of cold-water extraction and one step of hot-water extraction. Extraction 3: Three steps of hot-water extraction. Extraction 4: One step of cold-water extraction and three steps of hot-water extraction. Dried extract 1 (30 wt.% tannin), Liquid extract 2 (5 wt.% tannin), and Liquid extract 3 (5 wt.% tannin).

Table 5. Inventory data for the tannin extraction from spruce bark. Post-Extraction Scenario 1, 2, and 3 yielded dried extract 1, liquid extract 2, and liquid extract 3, respectively, in Figure 2. Table adapted from Ding et al. [20].

	Tannin Extraction	Description	Energy (kWh)
Extraction	Extraction	Extraction	22.80
Post-Extraction	Post-Extraction Scenario 1	Evaporation Spray Drying	211.1 7.600
	Post-Extraction Scenario 2	Evaporation	192.8
	Post-Extraction Scenario 3	Ultrafiltration Evaporation	0.410 97.50

3.1. Environmental Impact Assessment

Figure 3 shows the normalised environmental impacts of the various life cycle stages (A1–A3), including all scenarios within the system boundary (Figure 1). Normalising the different environmental impacts allowed for comparing the impacts on a standard scale to enable the identification of those impacts which are the most significant.

The raw material (A1) and transport (A2) stages contributed least to the various environmental impact categories. However, the plant cell culture-based biomanufacturing process (BMP, production stage A3) contributes significantly to the environmental impacts depending on the energy source employed. From a scenario analysis, the global warming potential was the most significant environmental impact, irrespective of the energy source used, followed by acidification. Using only solar- or geothermal-driven energy sources pose the most environmental impact in terms of global warming potential, acidification potential, and eutrophication even compared to the mixed energy scenario due to their higher embodied environmental factors (Table 2). In addition, using hydropower-derived energy sources produces the most negligible environmental impacts, followed by wind power. Comparing hydropower and wind sources, the wind source gives a higher carbon footprint than hydropower, while hydropower has a higher ozone depletion potential than wind. The energy mix scenario includes fossil fuel sources (natural gas, coal, and oil) accounting for 70% and renewable sources (wind, solar, hydropower and geothermal) constituting the remaining 30%. This gives a fair representation of Scotland's energy production, where the plant cell culture-based biomanufacturing process is located.

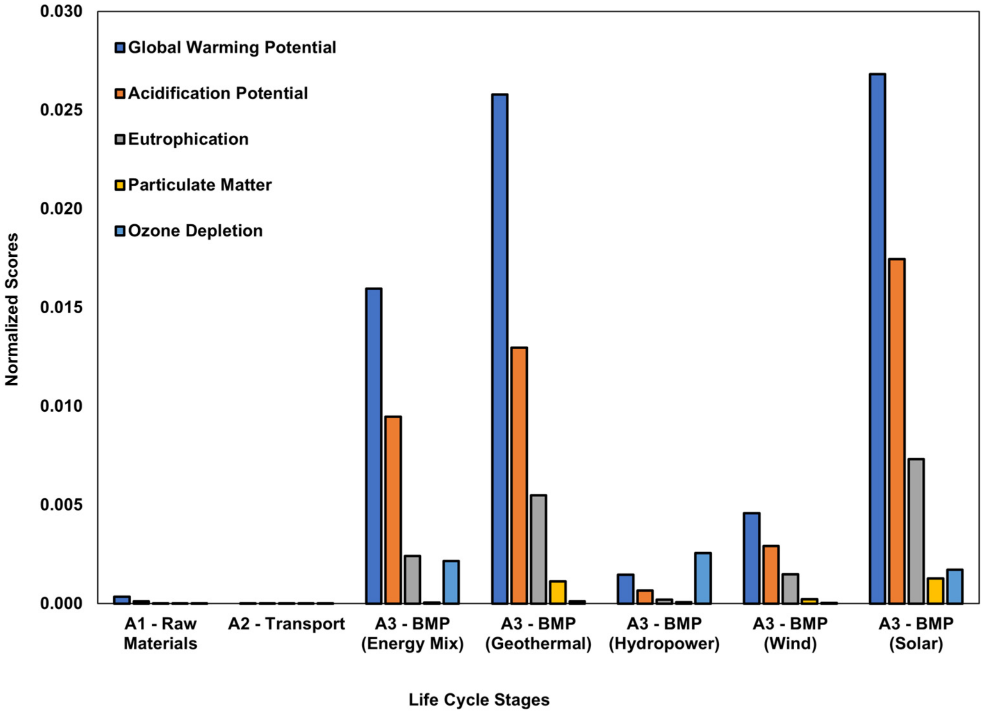

Figure 3. Normalized scores for the various environmental impact categories associated with GBL's biomanufacturing process across production stages A1–A3.

Figure 4 shows the environmental impacts of producing the various raw material inputs associated with the plant cell culture-based biomanufacturing process. The results indicate that Vitamin B5, Casein Hydrolysate, and PVP-10 production contribute significantly to global warming potential, due to their high embodied environmental factors (Table 4) and higher material input (Table 3). However, the materials' contribution to the biomanufacturing process's overall carbon footprint is insignificant.

Figure 5 shows the contribution of all stages, raw materials, transport, and the unit operations of the plant cell culture-based biomanufacturing process across the various environmental impact categories. The figure shows that the biomanufacturing stage contributes significantly to all impact categories. This could be attributed to the significant energy consumption associated with the biomanufacturing stage (2184 kWh).

3.2. Material Circularity Index (MCI) of the Biomanufacturing Process

The MCI for the biomanufacturing process was estimated to be 0.186, which indicates that the process has a relatively low material circularity, which can be attributed to two main reasons. Firstly, the raw materials used in the biomanufacturing process (currently) have 0% recycled or reused content (Table 6). This means that these process ingredients were made using virgin materials, which could include finite resources (Table 6). In addition, only 19% of the used media, regarded as waste, contains depleted ingredients, after the plant cell culture-based biomanufacturing process is recycled. Secondly, the product utility in terms of its longevity showed that the products have a life span (data provided by GBL) less than the industry average for comparable products, as shown in Table 7. A short product life span accelerates resource depletion and intensifies waste generation, negating circular economy principles. Prolonging product life enhances circularity by reducing the frequency of resource extraction and waste disposal. To improve the MCI of the biomanufacturing process, more sustainable practices, such as using raw materials with more recycled or reused contents, would be required. Additionally, using raw material inputs to the process that contain high proportions of reused and recycled materials and that are not entirely manufactured from virgin materials could improve the biomanufacturing process MCI. Improving the MCI of the biomanufacturing process would not only reduce its environmental impact but could also lead to cost savings and increased competitiveness in the market.

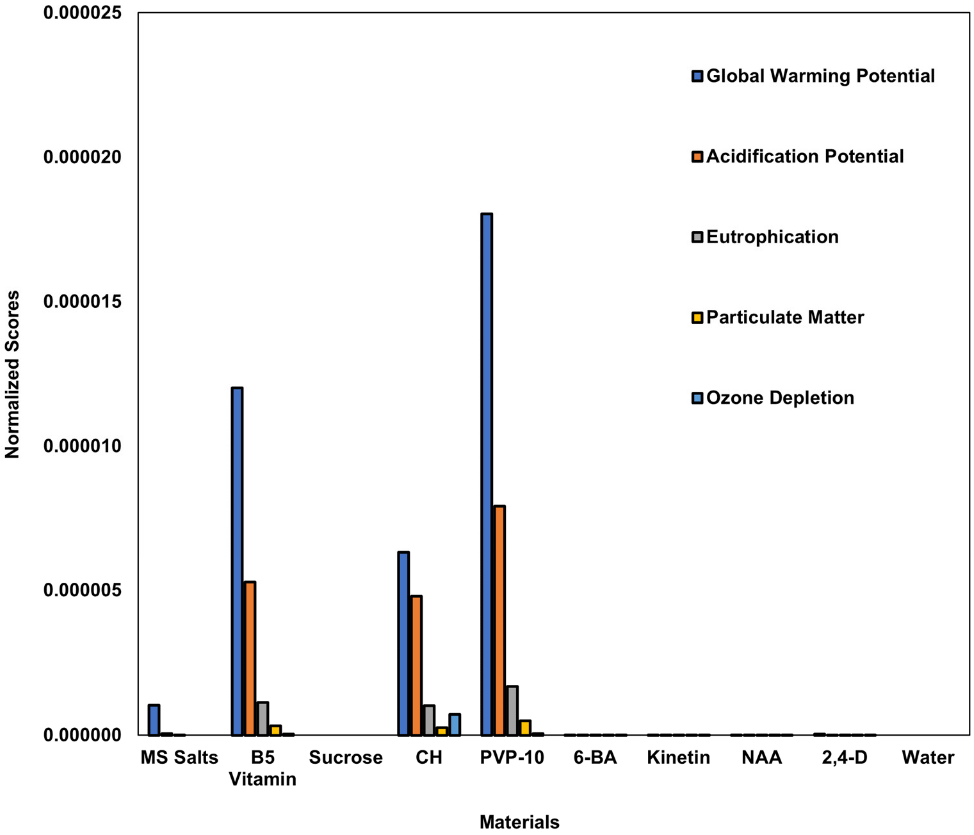

Figure 4. Environmental impacts associated with producing the various input materials associated with the plant cell culture based-biomanufacturing process.

Table 6. Recycled/reused content of raw materials used in extract production and recycled/reused fraction of waste generated.

Ingredients	Raw Materials		Waste	
	Recycled Content (%)	Reused Content (%)	Recycled Fraction (%)	Reused Fraction (%)
MS Salts	0	0	0.19	0
B5 Vitamin Stock 1000X	0	0	0.19	0
Sucrose	0	0	0.19	0
Casein hydrolysate	0	0	0.19	0
PVP-10	0	0	0.19	0
6-BA	0	0	0.19	0
Kinetin	0	0	0.19	0
NAA	0	0	0.19	0
2,4-D	0	0	0.19	0

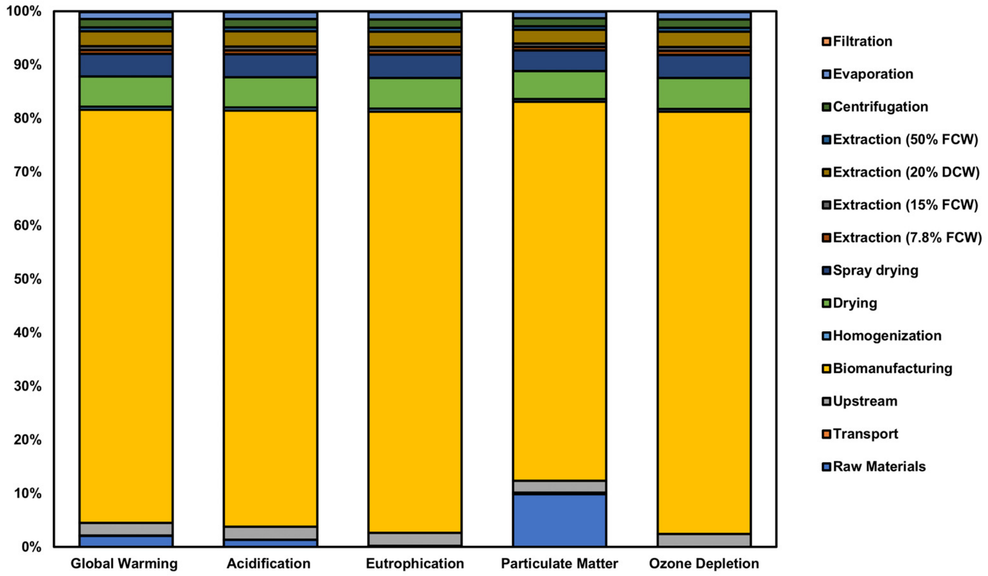

Figure 5. Contribution of the various stages of the life cycle of the plant cell culture-based biomanufacturing process to the environmental impact categories.

Table 7. Life span of the products obtained from the biomanufacturing process compared to the industry average lifespan.

Products (Output)	Life Span (Months)	Industry Average Life Span (Years)
Liquid extract [7.8, 25, 50% FCW]	24	2
Dried extract (Powder)	25	3
Liquid Extract 2 (20% DCW)	26	4
Evaporated extract (100X)	27	5

3.3. Biomanufacturing Process and Wild Harvest Approach

Figure 6 compares the plant cell culture-based biomanufacturing process with the tannin extraction process (wild harvest approach) based on the following environmental indicators: global warming potential, acidification potential, eutrophication, and ozone depletion potential. Compared with the wild harvest approach, the life cycle assessment (LCA) of the plant cell culture-based biomanufacturing process shows that the environmental impact categories are lower than those of the tannin extraction process across the life various cycle stages. The reason for this difference can be attributed to several factors. Firstly, the biomanufacturing process uses plant cell culture-based technology to produce natural ingredients, which is a sustainable approach. On the other hand, the tannin extraction process requires spruce bark, which involves land use and forest management practices. Secondly, the transport stage for the tannin extraction process is high due to the long distance for transportation of the spruce bark from Chile to the UK. This contributes significantly to the environmental impact categories associated with tannin extraction. Finally, the high electricity requirement for extracting the tannin from the spruce bark also contributes to its high-impact categories.

The plant cell culture-based biomanufacturing process uses renewable energy sources such as hydropower and wind power, which produce negligible environmental impacts. The extract production stage was compared using the same energy source (energy mix comprising 70% fossil fuel sources and 30% renewable sources). The LCA of the biomanufacturing process shows that it has lower impact categories than the tannin extraction process. This can be attributed to the sustainable and reliable approach. The sustainability of the approach stems from its lower environmental impact and the reliability of the shorter/local supply chains. Figure 7 compares the plant cell culture-based biomanufacturing process with the tannin extraction process (wild harvest approach) based on the different post-extraction scenarios using the global warming potential indicator.

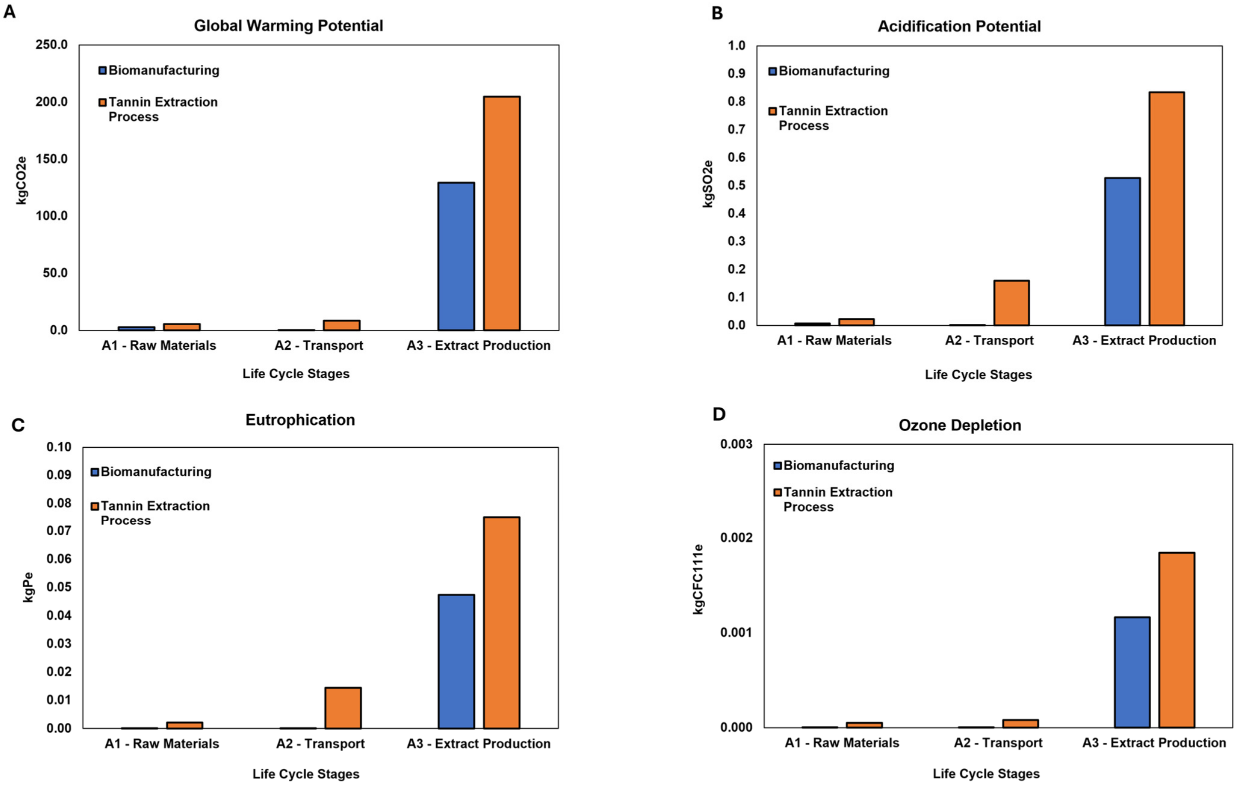

Figure 6. Environmental impact assessment for the biomanufacturing process and tannin extraction process (**A**) global warming potential, (**B**) acidification potential, (**C**) eutrophication, and (**D**) ozone depletion.

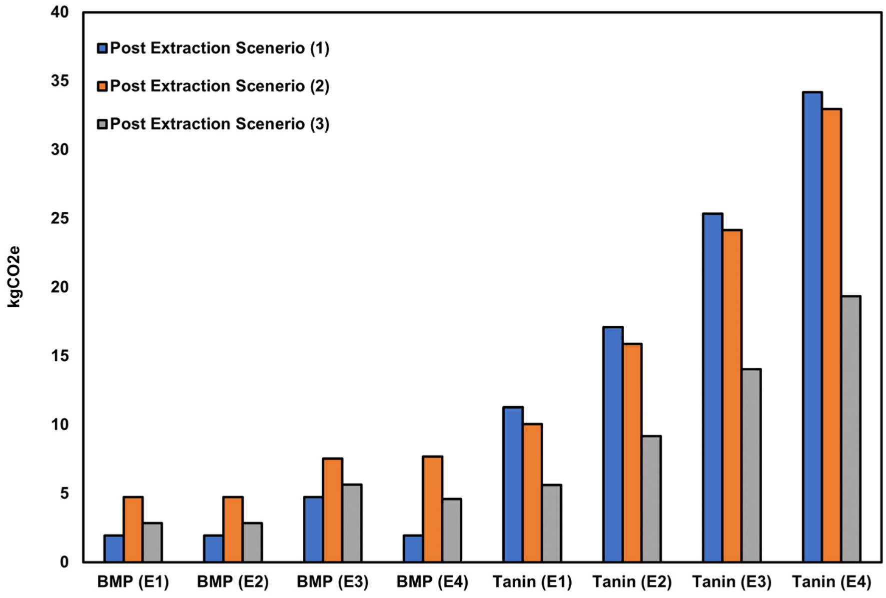

Figure 7. A comparison of GBL biomanufacturing vs. tannin extraction processes' greenhouse gas emissions impact (kgCO_2e) at different post-extraction scenarios. E1–E4 represents extraction scenarios for 7.8% FCW, 25% FCW, 50% FCW and 20% DCW in the case of biomanufacturing and dried extract 1, liquid extract 2 (5% tannin), and liquid extract 3 (5% tannin) for the tannin extraction process.

The greenhouse gas emissions footprint (kgCO_2e) of the tannin extraction from processed spruce bark was approximately 60% higher than that of the plant cell culture-based biomanufacturing process using electricity sourced across a variety of energy mixes. Irrespective of the post-extraction scenarios, the biomanufacturing process has a lower carbon emissions footprint than the tannin extraction process. Extracting tannins from spruce bark often involves energy-intensive processes, such as grinding, boiling, and chemical treatments, which require significant amounts of energy, water, and waste treatment. This can result in higher greenhouse gas emissions and increased energy consumption. The same trend exists regarding the post-extraction scenarios when considering other environmental impact categories—acidification, eutrophication, and ozone depletion. Figure 8 compares the post-extraction energy use for both processes. One of the reasons the energy requirements for the tannin extraction are higher than those for the biomanufacturing process is that the extraction procedure requires a significant amount of hot water for steam extraction of processed bark tissue.

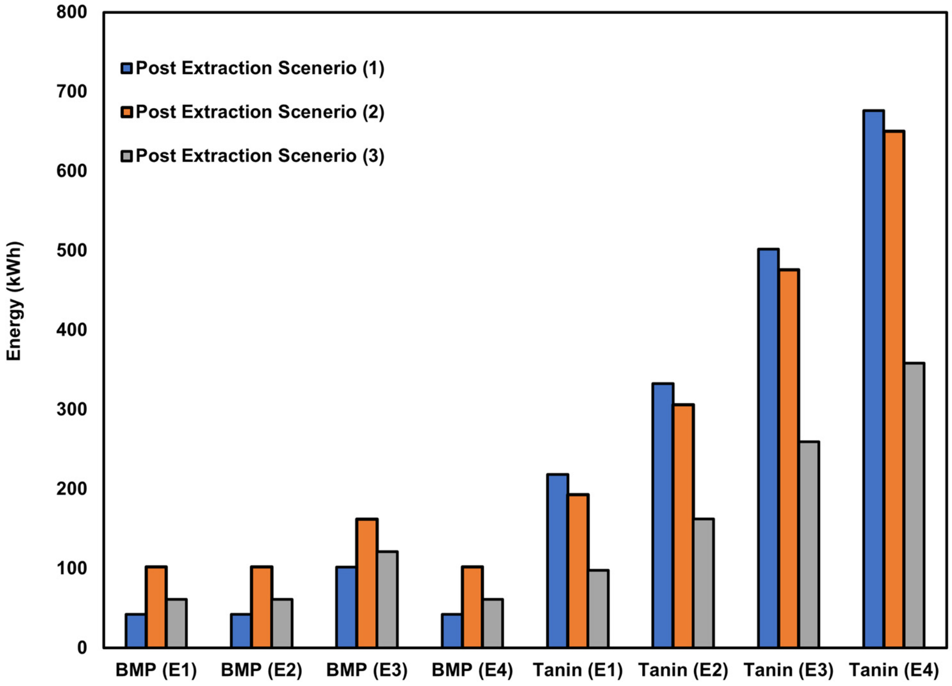

Figure 8. Energy requirements for the post-extraction scenarios for tannin extraction and biomanufacturing processes (BMP).

4. Conclusions

In pursuing sustainable industrial practices, this study undertook a Life Cycle Impact Assessment (LCIA) of a biomanufacturing process for producing plant natural product extracts. The system boundary of the analysis encompassed various production stages, including raw material sourcing, transportation, and production, focusing on understanding the environmental implications of the biomanufacturing process in order to suggest potential areas for process improvement.

4.1. Environmental Impact Insights

The results of the LCIA shed light on the nuanced environmental impacts associated with the biomanufacturing process. Unveiling the cradle-to-gate journey of plant extract production, the study identified key contributors to the plant cell culture-based biomanufacturing processes' environmental footprint. Notably, the production stage (A3) emerged as a significant contributor, primarily due to energy-intensive processes involved in the growth of plant cells and their subsequent extraction procedures. This underscores the importance of scrutinising energy consumption practices and energy sourcing choices in biomanufacturing.

4.2. Energy Source Influence

The study delved into the role of energy sources in shaping environmental impacts. Hydropower and wind power, being renewable sources, demonstrated the potential to minimize process environmental footprints. The comparative scenario analysis highlighted that the type of energy harnessed could substantially impact process outcomes, with associated global warming potential emerging as a consistent area of concern. The significance of this impact (while not unexpected per se) prompts a strategic evaluation of energy procurement practices, advocating for a more urgent transition toward the use of cleaner and more sustainable energy alternatives.

4.3. Material Circularity Index

The Material Circularity Index (MCI) assessment offered a unique lens into the current sustainability of the plant cell culture-based biomanufacturing process [27]). With an MCI of only 0.186, the process demonstrated a relatively low level of material circularity. This stemmed from the absence of recycled or reused content in the process raw material inputs and a product life span falling short of industry averages. This finding unveils pivotal areas to focus future process improvements, indicating the potential to enhance circularity by incorporating recycled or reused materials and extending the life span of the final product.

4.4. Comparative Analysis with the Wild Harvest Approach

A distinctive facet of this study lies in the comparative analysis with the wild harvest approach towards plant natural products manufacturing, specifically tannin extraction from spruce bark. The findings of this comparison unveil the sustainability advantages of the biomanufacturing process. The relative environmental friendliness of the plant cell culture-based technology, especially when coupled to the use of renewable energy sources, positions this approach as a less environmentally impactful alternative to traditional wild harvesting methods.

4.5. Recommendations for Improvement

The study concludes by offering actionable recommendations to enhance the future sustainability of plant cell culture-based biomanufacturing processes. Strategies include shifting toward cleaner energy sources, fostering circularity by incorporating recycled or reused materials, and exploring avenues to extend product life spans. Collaborative efforts with suppliers to ensure sustainable sourcing practices and a comprehensive evaluation of energy mix strategies in their production processes can further contribute towards improving environmental stewardship. As industries navigate the evolving sustainability landscape, this study lays the groundwork for future research into plant cell culture-based production processes. Further investigations could delve into optimising energy consumption in biomanufacturing, exploring innovative approaches to enhance circularity, and conducting more comprehensive life cycle assessments of alternative production methods. In conclusion, this study thoroughly examines the plant cell culture-based biomanufacturing process, offering valuable insights into its environmental impact and sustainability potential. By dissecting each production stage and conducting a comparative analysis, the study advances our understanding of the biomanufacturing landscape and charts a course for industry to adopt more sustainable operations. As we continue to grapple with the impacts of global climate change and resource depletion and availability challenges, endeavours such as these contribute to the broader narrative of fostering environmentally conscious industrial practices.

Author Contributions: Conceptualization, G.F.O. and D.M.; methodology, G.F.O. and R.O.A.; software, G.F.O. and R.O.A.; validation, G.F.O. and R.O.A.; formal analysis, G.F.O. and R.O.A.; investigation, G.F.O. and R.O.A.; resources, S.C.Z. and R.O.A.; data curation, R.O.A.; writing—original draft preparation, G.F.O. and R.O.A.; writing—review and editing, D.M., R.O.A. and Y.L.; visualization, G.F.O. and R.O.A.; supervision, G.F.O.; project administration, G.F.O.; funding acquisition, G.F.O. and D.M. All authors have read and agreed to the published version of the manuscript.

Funding: This research was funded by Scottish Funding Council (SFC) through the innovation voucher scheme.

Institutional Review Board Statement: Not applicable.

Informed Consent Statement: Not applicable.

Data Availability Statement: The datasets generated during and analysed during the current study are available from the corresponding author upon reasonable request.

Conflicts of Interest: Authors Samuel Casasola Zamora, Yuan Li and David McElroy were employed by the company Green Bioactives Limited. The remaining authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

References

1. Angelsen, A.; Jagger, P.; Babigumira, R.; Belcher, B.; Hogarth, N.J.; Bauch, S.; Börner, J.; Smith-Hall, C.; Wunder, S. Environmental income and rural livelihoods: A global-comparative analysis. *World Dev.* **2014**, *64*, S12–S28. [CrossRef] [PubMed]
2. Magrach, A.; Sanz, M.J. Environmental and social consequences of the increase in the demand for 'superfoods' world-wide. *People Nat.* **2020**, *2*, 267–278. [CrossRef]
3. Hudson, A.; Milliken, W.; Timberlake, J.; Giovannini, P.; Fijamo, V.; Massunde, J.; Chipanga, H.; Nivunga, M.; Ulian, T. Natural Plant Resources for Sustainable Development: Insights from Community Use in the Chimanimani Trans-Frontier Conservation Area, Mozambique. *Hum. Ecol.* **2020**, *48*, 55–67. [CrossRef]
4. Leão, T.C.C.; Lobo, D.; Scotson, L. Economic and Biological Conditions Influence the Sustainability of Harvest of Wild Animals and Plants in Developing Countries. *Ecol. Econ.* **2017**, *140*, 14–21. [CrossRef]
5. Zhao, J.; Hu, S.; Fan, L.; Zeng, Y.; Yang, Y.; Zhao, Y.; Lee, T.M. Does higher demand for medicinal plants lead to more harvest? Evidence from the dual trade of Nardostachy jatamansi and Fritillaria cirrhosa and Tibetan people's harvesting behavior. *Front. Ecol. Evol.* **2023**, *11*, 1145928. [CrossRef]
6. Zhao, J.; Fan, L.; Hu, S.; Poha; Jiayangji; Wan, A.K.Y.; Zeng, Y.; Yang, Y.; Duan, B.; Lee, T.M. Understanding indigenous people's traditional Chinese medicinal plants harvesting preferences to guide sustainable management. *People Nat.* **2023**, *5*, 1174–1186. [CrossRef]
7. Opuogulaya, R.; Gbosidom, V.L.; Ekiyor, T.H. Assessing the Sustainability of Thryonomys swinderianus Hunting in Ogoni Land. *Asian J. Biol.* **2021**, *13*, 1–8. [CrossRef]
8. Jaiswal, P.; Singh, A.V.; Yadav, V.K.; Kumar, A.; Kumari, N. Chapter 18—Phytochemical studies in the field of plant tissue culture. In *Advances in Plant Tissue Culture*; Chandra Rai, A., Kumar, A., Modi, A., Singh, M., Eds.; Academic Press: Cambridge, MA, USA, 2022; pp. 395–405. [CrossRef]
9. Jaiswal, N.; Verma, Y.; Misra, P. Elicitation enhanced the production of bioactive compound and biomass accumulation in callus cultures of *Glycyrrhiza glabra* L. *Vitr. Cell. Dev. Biol.-Plant* **2022**, *58*, 427–436. [CrossRef]
10. Nielsen, E.; Temporiti, M.E.E.; Cella, R. Improvement of phytochemical production by plant cells and organ culture and by genetic engineering. *Plant Cell Rep.* **2019**, *38*, 1199–1215. [CrossRef]
11. Patel, H.; Krishnamurthy, R. Elicitors in Plant Tissue Culture. *J. Pharmacogn. Phytochem.* **2013**, *2*, 60–65.
12. Rao, S.R.; Ravishankar, G.A. Plant cell cultures: Chemical factories of secondary metabolites. *Biotechnol. Adv.* **2002**, *20*, 101–153. [CrossRef] [PubMed]
13. Xu, J.; Zhang, N. On the way to commercializing plant cell culture platform for biopharmaceuticals: Present status and prospect. *Pharm. Bioprocess.* **2014**, *2*, 499–518. [CrossRef] [PubMed]
14. Bapat, V.A.; Kavi Kishor, P.B.; Jalaja, N.; Jain, S.M.; Penna, S. Plant Cell Cultures: Biofactories for the Production of Bioactive Compounds. *Agronomy* **2023**, *13*, 858. [CrossRef]
15. Rischer, H.; Szilvay, G.R.; Oksman-Caldentey, K.-M. Cellular agriculture—Industrial biotechnology for food and materials. *Curr. Opin. Biotechnol.* **2020**, *61*, 128–134. [CrossRef] [PubMed]
16. Häkkinen, S.T.; Nygren, H.; Nohynek, L.; Puupponen-Pimiä, R.; Heiniö, R.L.; Maiorova, N.; Rischer, H.; Ritala, A. Plant cell cultures as food-aspects of sustainability and safety. *Plant Cell Rep.* **2020**, *39*, 1655–1668. [CrossRef] [PubMed]
17. Nordlund, E.; Lille, M.; Silventoinen, P.; Nygren, H.; Seppänen-Laakso, T.; Mikkelson, A.; Aura, A.M.; Heiniö, R.L.; Nohynek, L.; Puupponen-Pimiä, R.; et al. Plant cells as food—A concept taking shape. *Food Res. Int.* **2018**, *107*, 297–305. [CrossRef] [PubMed]

18. Kobayashi, Y.; Kärkkäinen, E.; Häkkinen, S.T.; Nohynek, L.; Ritala, A.; Rischer, H.; Tuomisto, H.L. Life cycle assessment of plant cell cultures. *Sci. Total Environ.* **2022**, *808*, 151990. [CrossRef] [PubMed]
19. Curran, M.A. Life Cycle Assessment: A review of the methodology and its application to sustainability. *Curr. Opin. Chem. Eng.* **2013**, *2*, 273–277. [CrossRef]
20. Ding, T.; Bianchi, S.; Ganne-Chedeville, C.; Kilpelainen, P.; Haapala, A.; Raty, T. Life cycle assessment of tannin extraction from spruce bark. *Iforest-Biogeosci. For.* **2017**, *10*, 807–814. [CrossRef]
21. *ISO 14040: 2006*; Environmental Management—Life Cycle Assessment—Principles and Framework. International Organization for Standardization: Geneva, Switzerland, 2006.
22. ISO. *Environmental Management-Life Cycle Assessment-Requirements and Guidelines*; ISO: Geneva, Switzerland, 2006; Volume 14044.
23. Wernet, G.; Bauer, C.; Steubing, B.; Reinhard, J.; Moreno-Ruiz, E.; Weidema, B. The ecoinvent database version 3 (part I): Overview and methodology. *Int. J. Life Cycle Assess.* **2016**, *21*, 1218–1230. [CrossRef]
24. GreenDelta, G. *OpenLCA Software*; GreenDelta GmbH: Berlin, Germany, 2019.
25. Thévenot, A.; Rivera, J.L.; Wilfart, A.; Maillard, F.; Hassouna, M.; Senga-Kiesse, T.; Le Féon, S.; Aubin, J. Mealworm meal for animal feed: Environmental assessment and sensitivity analysis to guide future prospects. *J. Clean. Prod.* **2018**, *170*, 1260–1267. [CrossRef]
26. Rocchi, L.; Paolotti, L.; Cortina, C.; Fagioli, F.F.; Boggia, A. Measuring circularity: An application of modified Material Circularity Indicator to agricultural systems. *Agric. Food Econ.* **2021**, *9*, 9. [CrossRef]
27. Rigamonti, L.; Mancini, E. Life cycle assessment and circularity indicators. *Int. J. Life Cycle Assess.* **2021**, *26*, 1937–1942. [CrossRef]